REMARKS

Claims 3-7 and 12-17 have been cancelled.

Claims 1 and 2 have been amended to recite that the germline transmission efficiency in chicken chimeras is increased up to 49.7% by the present invention. Support for this amendment is on lines 18-21 of page 15 of the present specification.

Claim 1 has been amended to include the step (c) of injecting said cultured PGCs into a recipient embryo.

Claims 1 and 2 have been amended to contain the limitations described in claims 7, 12, 13, and 14.

Dependent claims 8 and 11 have been amended in accordance with cancelled claim 3.

Claims 8 and 11 have been amended to replace the expression "in vitro culture of PGCs" with "culturing PGCs in vitro."

No new matter has been added by the amendments.

The preceding amendments and the following remarks are believed to be fully responsive to the outstanding Office Action and are believed to place the application in condition for allowance.

The Examiner is respectfully requested to reconsider and withdraw the rejections in view of the above amendments and remarks as set forth hereunder.

I. Rejection under 35 USC §112

With regard to the rejection based on the enablement requirement, the Examiner has alleged that the specification does not enable transfecting PGC cells, maintaining them for at

least 5 days in culture, and using them to make germline chimeric avians as claimed.

According to the indication of the Examiner, claim 3, claiming a method for preparing a transgenic avian by injecting the transformed PGCs, has been cancelled.

Accordingly, the Applicant respectfully requests that this rejection be withdrawn.

With regard to the rejection concerning the breadth of avian, the Examiner has pointed out that claims 1-3 should be limited to chicken cells and producing chimeric chickens.

According to the indication of the Examiner, claims 1 and 2 have been amended to limit "avian" to "chicken."

Consequently, the Applicant respectfully requests that this rejection be withdrawn.

With regard to the rejection of indefiniteness, claim 1 has been amended to additionally include the limitation of step (c) of injecting said cultured PGCs into a recipient embryo. The construction of claim 1 as to how the method improves germline transmission efficiency of PGCs has been clarified by this amendment.

Claims 1 and 2 have been amended to recite that the germline transmission efficiency in chicken chimeras is increased up to 49.7% by the present invention. Thus, the metes and bounds of "improved germline transmission efficiency" in claims 1 and 2 have been clarified by the amendments.

In claim 2, the term "the improved germline tansmission efficiency" has been amended to "an improved germline transmission efficiency." By this amendment, claim 2 has been clarified.

In claims 8 and 11, the expression "in vitro culture of PGCs" has been amended to "culturing PGCs in vitro." Thus, claims 8 and 11 have been clarified by these amendments.

The rejection for indefiniteness relating to claims 3, 5, and 12-17 has been overcome by

cancelling these claims.

Accordingly, the Applicant respectfully requests that these rejections relating to indefiniteness be withdrawn.

II. Rejection under 35 USC §102

The Examiner rejected claims 1-14 as being anticipated by the cited references.

The presently claimed invention relates to a novel idea of in vitro culturing chicken PGCs at least ten days in order to obtain a great improvement in germline transmission efficiency in chicken chimeras.

The amended instant claims have the characteristics of i) germline transmission efficiency of up to 49.7%, ii) culturing PGCs in vitro at least 10 days, iii) *in vitro* cultured PGCs express SSEA-1, iv) injecting cultured PGCs into the recipient embryo is carried out by injecting PGCs into the dorsal aorta of the recipient embryo, and v) culturing PGCs *in vitro* is conducted on a gonadal stroma feeder cell layer.

The reference of Cell Biology International (1997, Vol. 21, No. 8, pg 495-499) by Chang disclosed that gPGCs from chicken embryonic germinal ridges are cultured in vitro for 5 days (*Preparation and culture of gPGCs* in page 496), and not at least 10 days, as is specified in the present claims. In addition, the reference does not disclose whether gPGCs express SSEA-1 or not. Furthermore, the reference does not teach or suggest the improvement of germinal transmission efficiency in the chimera by the process of injecting the PGCs *in vitro* cultured at least ten days into the recipient embryo.

The reference of Mol. Reproduction and Development (2000, Vol. 56, pg 475-482) by

Park disclosed that they used EG cells after three or four passages to produce chimeras (Production of Somatic Chimeras and PCR analysis in page 479) and the chicken EG cells were plated and cultured on mitotically active chicken embryonic fibroblasts (CEFs) (Culture of the Chicken EG cells in page 477). It is noteworthy that the PGCs of the present invention are cultured on gonadal stroma cells as a feeder cell layer. In addition, the reference of Park does not also teach or suggest the idea of the improvement of germinal transmission efficiency in the chimera of up to 49.7% by the process of injecting PGCs *in vitro* cultured at least ten days into the recipient embryo.

The reference of Transgenic Research (February 2002, Vol. 11, No. 1, pp. 85; presented to the public at the Transgenic Animal Research Conference) by Kim does not disclose the specific conditions of *in vitro* culturing PGCs. That is, there is no description as to the feeder cell layer used in *in vitro* culturing PGCs and the expression of SSEA-1 in the cultured PGCs.

The reference of Transgenic Research (February 2002, Vol. 11, No. 1, pp. 85; presented to the public at the Transgenic Animal Research Conference) by Zandong discloses transfecting cultured chicken embryonic fibroblasts (CEFs) rather than PGCs with constructed vector. In addition, the reference of Zandong does not disclose injecting PGCs into chicken embryo. Furthermore, there is no description as to the improvement of germinal transmission efficiency in the chimera by the process of injecting *in vitro* cultured PGCs at least ten days into the recipient embryo.

The reference of Theriogenology (Nov. 2002, Vol. 58, pg 1531-1539) by Han does not disclose that the improvement of germinal transmission efficiency of up to 49.7% by injecting the PGCs *in vitro* cultured at least ten days into the recipient embryo.

The references indicated by the Examiner do not disclose the technical features of the present invention. Hence, contrary to the Examiner's allegation, a skilled person would have had no reasonable expectation that PGCs originated from embryonic gonad and cultured for at least ten days could successfully used for improving germline transmission, as recited in the present claims.

We would therefore ask that the Examiner's anticipation rejection be withdrawn.

CONCLUSION

In summary, in order to overcome the rejection of enablement requirement, claim 3 has been cancelled. The rejection relating to the breadth of avian has been overcome by limiting "avian" to "chicken" in claims 1 and 2. With regard to the rejections of indefiniteness, the rejections have been overcome by the amendments of claims 1, 2, 8, and 11, and cancellation of claims 3, 5-7, and 12-17.

With regard to the rejection relating to anticipation by the cited references, the references indicated by the Examiner do not teach or suggest the technical features of the present invention. Accordingly, a skilled person would have had no reasonable expectation that PGCs cultured for at least ten days could successfully used for improving germline transmission.

Therefore, in view of the foregoing amendments and remarks, the Applicant respectfully requests reconsideration of the presently claimed invention and the timely allowance of the pending claims.

If there are any charges not covered or any credits, please apply them to Deposit Account No. 03-2095.

Respectfully submitted,

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